

AD-A269 620GE

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nd Reports, 1215 Jefferson Davis Highway, Suite 1204, Arlington, VA 22202-4302, and
03.

1. Agency Use Only (Leave blank).

1993

3. Report Type and Dates Covered.
Final - Proceedings

4. Title and Subtitle.

An Evaluation of Titanium Exposed to Thermophilic and Marine Biofilms

5. Funding Numbers.
Contract

Program Element No. 0601153N

Project No. 03103

Task No. 320

Accession No. DN094463

Work Unit No. 13332A, 13331A

6. Author(s).

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7. Performing Organization Name(s) and Address(es).

Naval Research Laboratory
Ocean Sciences Branch
Stennis Space Center, MS 39529-50048. Performing Organization
Report Number.

NRL/PP/7333--92-0003

9. Sponsoring/Monitoring Agency Name(s) and Address(es).

Naval Research Laboratory
Operations Research and Strategic Planning
Stennis Space Center, MS 39529-500410. Sponsoring/Monitoring Agency
Report Number.

NRL/PP/7333--92-0003

11. Supplementary Notes.

Published in Corrosion 93.

DTIC
ELECTE
SEP 23 1993
S A D

12a. Distribution/Availability Statement.

Approved for public release; distribution is unlimited.

12b. Distribution Code.

13. Abstract (Maximum 200 words).

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93-22073

93 9 22 03 6



10p8

14. Subject Terms.

Titanium, microbiologically influenced corrosion, extreme environment

15. Number of Pages.

10

16. Price Code.

17. Security Classification
of Report.
Unclassified18. Security Classification
of This Page.
Unclassified19. Security Classification
of Abstract.
Unclassified20. Limitation of Abstract.
SAR



AN EVALUATION OF TITANIUM EXPOSED TO THERMOPHILIC
AND MARINE BIOFILMS

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ABSTRACT

Flat coupons of Grade 2 titanium were immune to microbiologically influenced corrosion in high temperature environments (55–70°C) containing sulfur-oxidizing (SOB) and sulfate-reducing bacteria (SRB). SMO 254 was heavily pitted after similar exposure to thermophilic SRB. Hydrides did not form in titanium weld regions exposed to thermophilic hydrogen-producing bacteria. After a one-year exposure to Pacific Ocean seawater, grade 2 titanium and SMO 254 were colonized by diatoms and other algal species. E_{corr} increased continuously for the stainless steel, but not for titanium. The latter result can be explained by the poor catalytic activity of the oxide layer on titanium for oxygen reduction. The capacitance obtained with electrochemical impedance spectroscopy (EIS) remained constant for both metals with titanium showing values close to the theoretical value normally observed only for inert materials. The high values of the polarization resistance (R_p) and the capacitive nature of the impedance demonstrate that the materials remained in the passive state throughout the entire exposure period.

Keywords: titanium, microbiologically influenced corrosion, extreme environment

INTRODUCTION

The resistance of titanium alloys to microbiologically influenced corrosion (MIC) in operating systems and laboratory experiments has been repeatedly demonstrated.^{1–5} Corrosion resistance is derived from the formation of a highly stable, adherent protective titanium dioxide surface film typically 50–200 Å in thickness. The TiO_2 film can be either highly crystalline, rutile and/or somewhat amorphous anatase.¹ Titanium metal is highly reactive and has a high affinity for oxygen, so that protective oxides form spontaneously when metal is exposed to trace amounts of oxygen. TiO_2 is stable over a wide range of redox potentials and pH values, including those produced by mesophilic bacteria within biofilms.⁶ Stable film formation is favored under mildly reducing to highly-oxidizing conditions. Welded titanium alloys reportedly exhibit the same basic range of corrosion resistance as wrought metal forms.¹

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The resistance of metals to corrosion is strongly influenced by temperature. The minimum temperature for crevice corrosion in titanium alloys is 75°C.⁷ Titanium alloys are fully resistant to reduced chemical species associated with anaerobic microbial activity including ammonia, sulfides, hydrogen sulfide, nitrites, ferrous ions and organosulfur compounds at temperatures up to 100°C. Titanium oxide films remain intact under anaerobic reducing conditions down to pH=2 at 100°C. Titanium alloys are also resistant to absorption of hydrogen produced by bacteria at temperatures below 75–80°C where the diffusion coefficient for hydrogen in alpha titanium is negligible.¹

Experiments were designed to evaluate the corrosion resistance of titanium and a 6% molybdenum-containing stainless steel at high temperatures in the presence of SOB and SRB. Titanium hydride formation at titanium welds was evaluated at thermophilic temperatures in the presence of hydrogen-producing bacteria. Results of long-term exposures of the materials to natural seawater were also evaluated.

METHODS AND MATERIALS

Long-Term Exposure to Seawater

The experimental design for long-term exposures of titanium and a 6% Mo-containing stainless steel alloy in a seawater environment at the Naval Civil Engineering Laboratory in Port Hueneme, CA has been described elsewhere.^{8–11} Triplicate specimens were exposed for each material. The impact of marine biofilm formation was monitored using E_{corr} and EIS. Double-layer capacitance (C_{DL}) and R_p were calculated for the materials as a function of exposure time.

Exposures to Thermophilic Bacteria

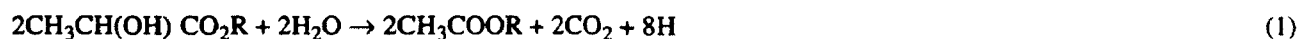
Maintenance of cultures and electrochemical experiments were conducted in bacterial growth media specific for each organism.

Sulfur-Oxidizing Bacteria

The alloys were exposed at 70°C for 84 days in a culture medium (ATCC 1723) containing *Sulfolobus acidocaldarius* (ATCC 33909), an aerobic, gram-negative, sulfur-oxidizing organism. The medium was adjusted to a pH=3.0 and filter sterilized before inoculation. *S. acidocaldarius* is an acidophilic, facultative autotroph that can use elemental sulfur as an energy source. Optimal growth range is 70–75°C at pH 2–3.

Sulfate-Reducing Bacteria

A mixed culture of facultative anaerobic bacteria known to contain SRB was isolated from a corrosion failure on a surface ship from a carbon steel waster piece and has been described elsewhere.¹² Because the mixture of organisms was shown to be particularly aggressive to some alloys at mesophilic temperatures,^{12,13} the culture was gradually acclimated to 55°C so that corrosion influenced by the culture could be evaluated at a higher temperature. After 2 weeks organisms grew at the elevated temperature in Postgate B medium¹⁴ to which 25g/l NaCl was added. Lactate was the electron donor. Typical metabolic reactions for SRB using lactate as a carbon source in a sulfate medium can be described as follows:



N_2 was bubbled through the medium during the experiments.

Hydrogen-Producing Bacteria

The thermophilic, hydrogen-producing bacterium, *Clostridium thermosaccharolyticum* (ATCC # 31960) is a saccharolytic bacterium capable of producing large quantities of hydrogen gas from fermentation of sugars. The organism was maintained in basal thermophile medium (ATCC #1465) at 62°C. The primary metabolites are butyric and acetic acids with considerable amounts of ethanol formed by resting cells during glucose fermentation. In its simplest form the fermentation process can be described as follows:¹⁵



Prior to each experiment, a welded strip of Grade 2 titanium was added to an autoclaved cell purged with oxygen-free gas: 20% CO₂/80% N₂. Inoculated culture medium was added anaerobically to the cell with a reducing agent (Na₂S) to stabilize the redox potential at an optimum level for the bacteria.

Electrochemical Cell

The corrosion behaviors of titanium and SMO 254 were measured in dual cell corrosion-measuring devices in the presence of thermophilic SOB and SRB. The dual- or split-cell allows continuous monitoring of changes in the corrosion rate. In this technique, two identical electrochemical cells are biologically separated by a semipermeable membrane. The two working electrodes are connected to a zero resistance ammeter. Bacteria are added to one of the two cells and the resulting galvanic current is recorded on a strip chart recorder. The sign and magnitude of the galvanic current can be used to determine details of the corrosive action of the bacteria. In the absence of any perturbation by the bacteria, the galvanic current would remain at zero. Using this technique, inherent differences between the electrochemical properties of titanium and the two stainless steels will be nullified and only the contribution of bacteria to corrosion processes will be measured. Polarization curves were recorded at intervals for both electrodes in the dual-cell in a potential range of $E_{\text{corr}} \pm 30$ mV and analyzed using POLFIT software¹⁶ to determine values for b_a , b_c , B , i_{corr} , R_p , and E_{corr} .

At the conclusion of the experiments, all electrodes were evaluated for weight loss and surfaces examined using energy dispersive spectroscopy (EDS) coupled with an environmental scanning electron microscope (ESEM) that allows one to image wet, non-conducting material, including biofilms, without fixation or drying.¹⁷ All surfaces were documented with photomicrographs and EDS spectra. Titanium hydride formation was evaluated microstructurally and by direct hydride analysis.

RESULTS AND DISCUSSION

The composition of SMO 254 is presented in Table 1. Grade 2 titanium is commercially pure.

Long Term Exposure to Seawater

After a one year exposure to flowing Pacific Ocean seawater, E_{corr} values for titanium and the 6% Mo stainless steel were shifted in the positive direction (Figure 1). E_{corr} values are the average of three measurements taken from replicate specimens. Johnsen and Bardal¹⁸ reported ennoblement of SMO 254 after exposure to natural seawater and indicated that the dramatic increase of E_{corr} in their measurements as a result of biofilm formation might lead to an increased susceptibility for localized corrosion. In the Pacific Ocean exposures, all surfaces remained shiny and free of localized corrosion. Macroscopically and microscopically, surfaces were colonized with diatoms and filamentous algae (Figure 2 a, b). The increase of E_{corr} with time shown in Figure 1 is in agreement with earlier results of Little, Mansfeld and co-workers conducted at the same test site.⁸⁻¹¹ While ennoblement of E_{corr} for stainless steels in natural seawater may be a common phenomenon, the degree of ennoblement appears to depend on seawater properties at the exposure site as demonstrated by the slow rate of SMO 254 ennoblement observed at Fort Huencine, CA. Ennoblement of E_{corr} is usually considered to be due to an increase in the rate of the oxygen reduction reaction by a still unknown mechanism.¹⁹ Based on this

assumption it is easy to understand the decreased ennoblement of titanium (Figure 1) which is a very poor catalyst for the oxygen reduction reaction. This result and the documented resistance of titanium to localized corrosion make it unlikely that localized corrosion will occur in natural seawater under ambient conditions. R_p increased markedly for titanium from day 13 to experiment conclusion, indicating a decrease in corrosion rate (Figure 3). The reverse was true of R_p for SMO 254, i.e., R_p decreased, indicating an increase in corrosion rate (Figure 3). C_{DL} for the metals remained essentially unchanged over the experimental period even though surfaces had been colonized by diatoms and other algae (Figure 4). C_{DL} for titanium was lower than that of the stainless steel and remained close to the theoretical value for the double layer capacitance between 20 and 30 $\mu\text{F}/\text{cm}^2$ (Figure 4), usually observed only for inert metals such as Pt.

Sulfur-Oxidizing Bacteria

Microorganisms colonized all surfaces exposed to the thermophilic SOB culture (Figure 5 a, b). Galvanic corrosion current measurements between biotic and abiotic cells remained below detection limits throughout the experiment and no significant weight loss was measured for either material over the 84 day exposure (Table 2). Inspection of the data for titanium in Table 2 shows that pH stayed around 3 for the abiotic solution, but increased to values higher than 4 for the biotic solution. E_{corr} was ennobled after exposure for 30 and 84 days. The increase in pH in the biotic solution was accompanied by a lower value of E_{corr} , suggesting that E_{corr} is influenced by the reversible potential for the oxygen reduction reaction on a passive surface. In the dual cell experiments the electrode in the biotic solution containing the thermophilic SOB culture was the anode due to the more positive E_{corr} in the abiotic solution. No localized corrosion was observed by ESEM/EDS.

Sulfate-Reducing Bacteria

Surfaces of titanium and SMO 254 exposed to the high-temperature-acclimated SRB were covered with bacterial cells after the 167 day exposure. Galvanic corrosion remained below detection limits throughout the experiment. Inspection of the data in Table 3 for titanium shows that for biotic solutions pH decreased slightly from the initial value at $t = 0$. A sharper drop of pH with exposure time occurred in the abiotic solution. A trend for more positive E_{corr} values at lower pH can be observed in Table 3. For titanium E_{corr} is more positive than the initial value. The low i_{corr} values and low corrosion rates determined from weight loss data (Table 3) are an indication that titanium remained passive throughout the test. Based on these results it can be concluded that colonization of titanium with SRB did not produce significant changes in corrosion behavior. A corrosion rate of 0.2 mpy was calculated for SMO 254 exposed to SRB at 55°C. E_{corr} values for SMO 254 exposed to SRB were decidedly more negative than their abiotic counterparts, a further indication of electroactive surfaces. ESEM micrographs document pitting in SMO 254 and the absence of localized corrosion in titanium (Figure 6 a, b).

Numerous reports document MIC in stainless steel alloys containing 2–3% molybdenum.^{20–24} Scott and Davies reported microbial attack of 904L stainless steel containing 4–5% molybdenum. Examination of pitting failures of 904L tube heat exchangers indicated the presence of many bacteria, including SRB, in the cooling water, on surfaces and in areas of corrosion.²⁵ There are two reports in the literature of MIC of SMO 254 in the presence of mesophilic iron-oxidizing bacteria in laboratory experiments.^{6,26}

Hydrogen-Producing Bacteria

Surfaces of the titanium weld materials exposed to *C. thermosaccharolyticum* were colonized with bacterial cells (Figure 7). Hydrogen analyses results were as follows: unexposed controls—base metal, 36 ppm and weld metal, 37 ppm; exposed—base metal, 45 ppm and weld metal, 37 ppm. Microphotographs of the exposed base and weld samples showed no evidence of hydride formation (Figure 8 a, b). Based on measured hydrogen concentrations there may have been a slight absorption of hydrogen after exposure to the thermophilic hydrogen-producing bacterium. Increased hydrogen after high temperature aqueous exposure may be due to the growth of an oxide film by breakdown of water.

CONCLUSIONS

Titanium had low corrosion rates under the exposure conditions with no localized attack. No evidence of hydride formation was observed when a titanium weld was exposed to a thermophilic hydrogen-producing bacterium. SMO 254 was pitted and had an increased corrosion rate in the presence of SRB maintained at 55°C. E_{corr} for the 6% Mo stainless steel shifted gradually in the positive direction during the one-year exposure to Pacific Ocean seawater. This increase occurred at a slower rate than reported for other locations. It can be concluded that ennoblement of E_{corr} for stainless steels is a common phenomenon in natural seawater. However, the rate of the ennoblement appears to depend on the properties of the seawater at a given exposure site. E_{corr} for titanium was not ennobled. This result has been explained by the poor catalytic activity of the titanium oxide layer which does not allow the increase of the rate of oxygen reduction with time, leading to ennoblement as observed for the stainless steels. The capacitive nature of the impedance spectra and the high values of R_p are another indication that the two materials remained passive throughout the one-year exposure time without initiation of localized corrosion. The lack of an ennoblement of E_{corr} for titanium and its resistance to localized corrosion make it unlikely that titanium will suffer from MIC in seawater at ambient conditions.

ACKNOWLEDGMENTS

This work was supported by TIMET (Titanium Metals Corp.), West Lake Mead Dr., Henderson, NV 89015. NRL Contribution number NRL/PP/7333-92-0003.

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TABLE 1
COMPOSITION OF MATERIALS, WEIGHT %

Materials	C	Mn	Si	Cr	Ni	P	S	Mo	Cu	N	Ti	Fe
Ti (grade 2)											CP*	
SMO 254	0.02	1.00	0.80	19.5–20.5	17.5–18.5	0.030	0.010	6.0–6.5	0.5–1.0	0.18–0.22		51.42–54.46

*Commercially Pure

TABLE 2
ELECTROCHEMICAL PARAMETERS AFTER EXPOSURE TO THERMOPHILIC SOB

Material	Exposure Time (Days)	pH	Galvanic Current	b _a (mV)	b _c (mV)	B (mV)	i _{corr} (μA/cm ²)	R _p (10 ⁵ ohm • cm ²)	E _{corr} (mV)	Correlation Coefficient	Corrosion Rate from Weight Loss (mpy)
Ti	0	3.00							-207		
abiotic	30	3.22	*	1	25	0.4	0.001	6.65	297	.789	
biotic	30	4.28				**			141		
abiotic	84	3.05	*	60	88	15.6	0.011	11.60	236	.786	0.0
biotic	84	4.71		3	22	1.1	0.003	3.52	151	.603	0.0
SMO 254	0	3.00							81		
abiotic	30	2.95	*	43	24	6.6	0.007	9.10	237	.998	
biotic	30	3.89		20	19	4.2	0.007	5.90	84	.755	
abiotic	84	2.88	*	40	18	5.3	0.002	20.96	207	.992	0.0
biotic	84					**			85		0.0

* below detection limits

** results below calculation limits

TABLE 3
ELECTROCHEMICAL PARAMETERS AFTER EXPOSURE TO THERMOPHILIC SRB

Material	Exposure Time (Days)	pH	Galvanic Current	b _a (mV)	b _c (mV)	B (mV)	i _{corr} (μA/cm ²)	R _p (10 ⁵ ohm • cm ²)	E _{corr} (mV)	Correlation Coefficient	Corrosion Rate from Weight Loss (mpy)
Ti	0	5.71	*						-202		
abiotic	35	3.77	*	47	24	6.8	0.010	6.56	122	.998	
biotic	35	4.57	*	68	11	4.2	0.015	2.54	100	.993	
abiotic	98	3.89	*	35	18	5.2	0.002	25.60	147	.992	
biotic	98	4.66		78	10	3.8	0.016	2.54	107	.996	
abiotic	167	3.91	*	12	24	3.4	0.003	11.36	185	.884	0.0
biotic	167	4.65		39	11	3.7	0.003	11.90	105	.989	0.0
SMO 254	0	5.71	*						-178		
abiotic	35	3.92	*	54	14	4.7	0.012	3.59	124	.992	
biotic	35	4.61	*	93	24	8.2	5.520	0.01	-333	.999	
abiotic	98	4.10	*	46	19	5.9	0.005	11.97	199	.994	
biotic	98	4.73		31	27	6.3	0.811	0.08	-225	.992	
abiotic	167	4.04	*	44	20	6.1	0.012	5.05	72	.995	0.0
biotic	167	4.64				**			-267		0.2

* below detection limits

** results below calculation limits

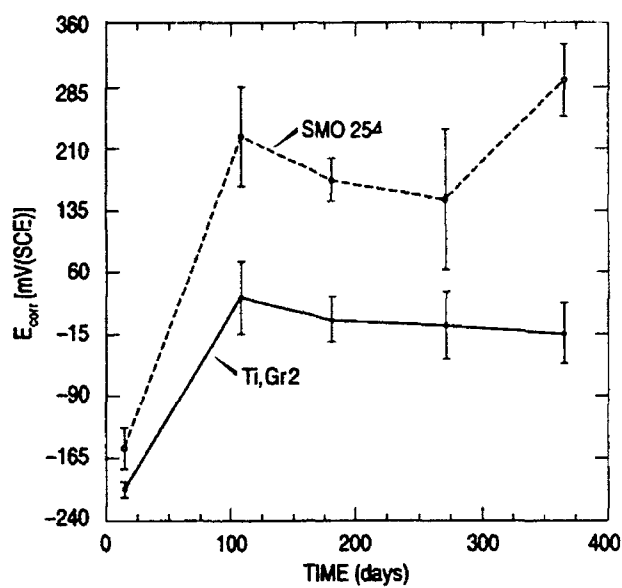


FIGURE 1 – E_{corr} after exposure to Pacific Ocean seawater

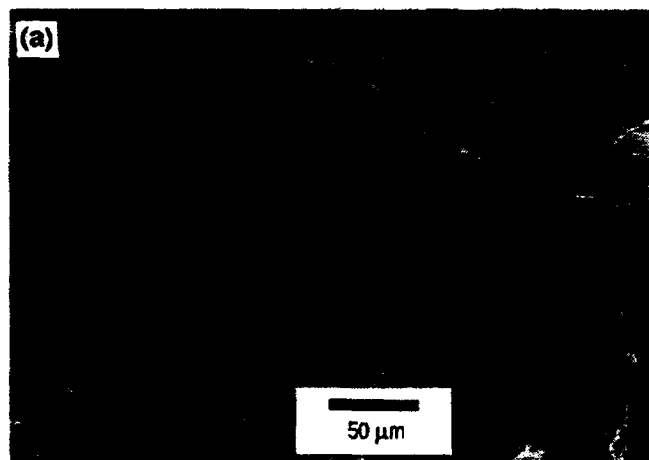
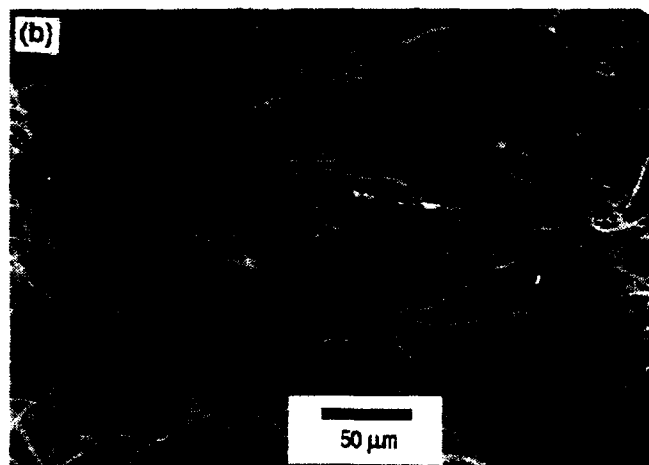


FIGURE 2 – Metal surfaces after 1 year exposure to Pacific Ocean Seawater (a) Ti and (b) SMO 254

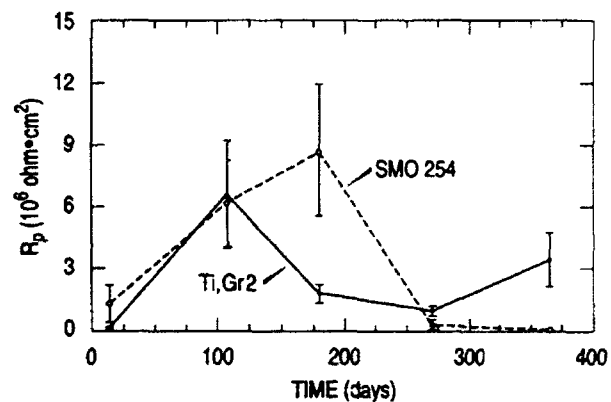


FIGURE 3 – Polarization resistance after exposure to Pacific Ocean seawater

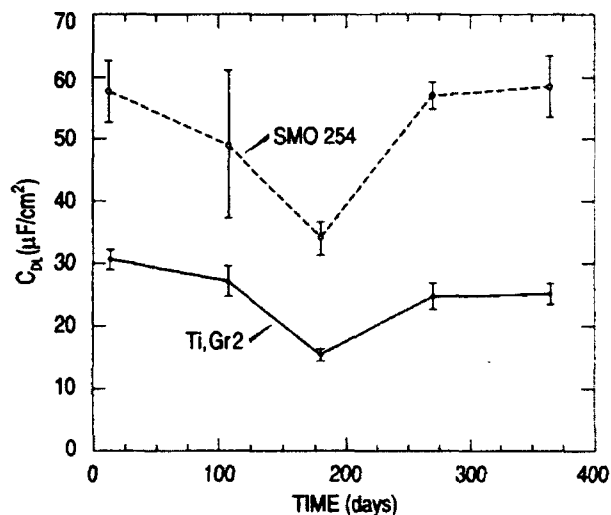


FIGURE 4 – Electrode capacitance (C_{dc}) after exposure to Pacific Ocean seawater



FIGURE 5 – Metal surfaces after 84 days exposure to SOB at 70°C (a) Ti and (b) SMO 254

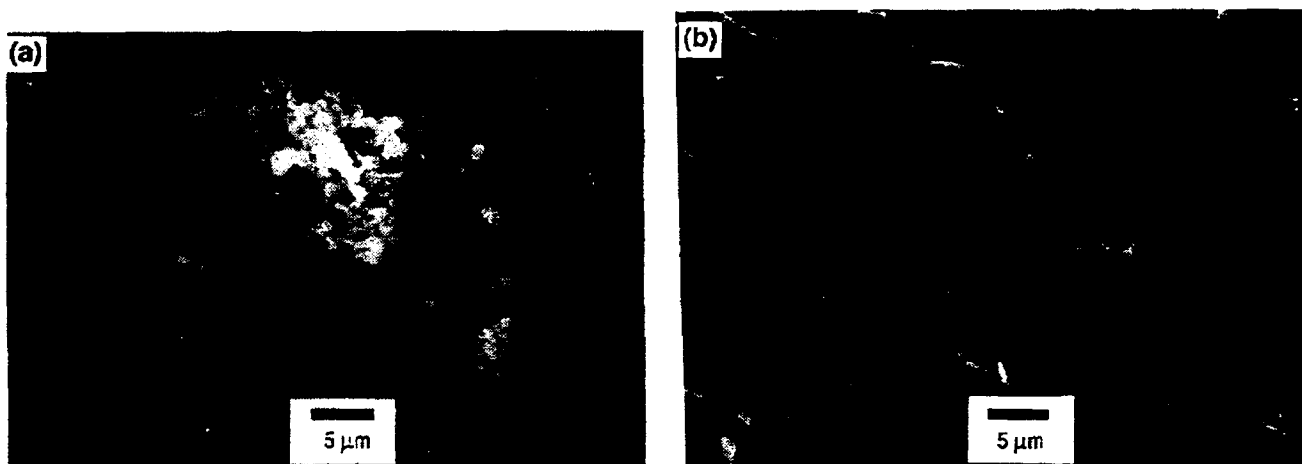


FIGURE 6 – Metal surfaces after 167 days exposure to SRB at 55°C (a) Ti and (b) SMO 254

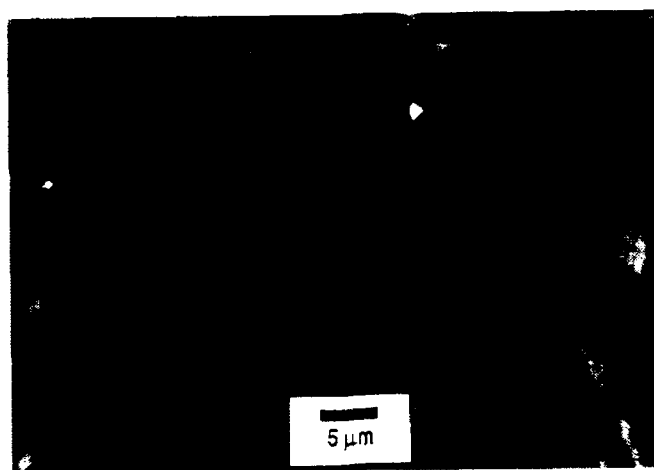


FIGURE 7 – Titanium weld colonized by hydrogen-producing bacteria

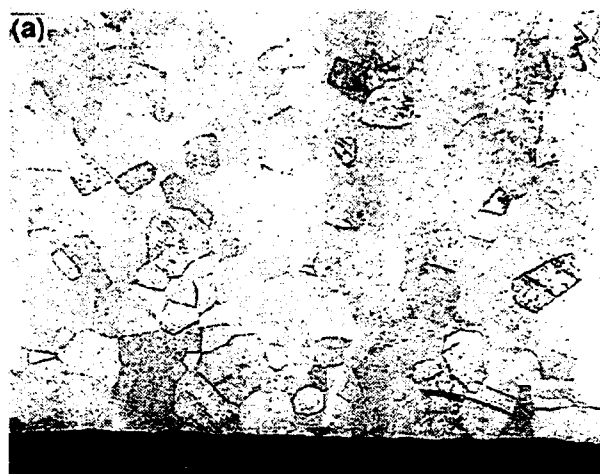


FIGURE 8 – Titanium exposed to *C. saccharolyticum*
(a) base metal and (b) weld